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Research Article

Modelling disease spread in real landscapes: Squirrelpox spread in Southern Scotland as a case study

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Abstract

There is increasing evidence that invading species can gain an advantage over native species by introducing novel disease. A clear understanding of the role of disease in the expansion of introduced and invading species is therefore essential for the conservation of native species. In this study we focus on the case study system of the UK red and grey squirrel system in which disease-mediated competition has facilitated the replacement of red squirrels by greys. We modify a deterministic model of the squirrel system in which the competition and infection dynamics are well understood to produce a stochastic model which includes a realistic representation of the heterogeneous habitat in Southern Scotland. The model is used to examine the potential spread of infection (squirrelpox virus) through the squirrel system and to examine the impact of conservation measures that control grey squirrel numbers in an attempt to contain disease spread. The results have direct implications for conservation management and we discuss how they have helped shape current and future policy for red squirrel conservation in Scotland. The methods in this study can be readily adapted to represent different systems and since the stochastic population and disease dynamics are underpinned by classical deterministic modelling frameworks the results are applicable in general.

Introduction

The introduction and invasion of exotic organisms has caused catastrophic damage to native communities (Kolar and Lodge, 2001) with the invasion and establishment of non-native species recognised as a major international threat to native biodiversity (DEFRA, 2007; Ehrenfeld, 2011; Simberloff, 2011). Therefore, understanding the mechanisms and processes that determine successful invasion are key challenges in ecological theory that seek to underpin conservation efforts and thereby sustain ecosystems (Manchester and Bullock, 2000; Sutherland et al., 2006). There are likely to be many factors that determine the likelihood and rate of spread of invasive species (Kolar and Lodge, 2001) but it is now recognised that shared infectious disease is a key determinant of invasive success (Daszak et al., 2000; Prenter et al., 2004; Strauss et al., 2012; Dunn and Hatcher, 2015; Vilcinskas, 2015).

One aspect that is gaining increasing attention is where the invasive species gains an advantage by introducing a novel disease to the native system. There is evidence that parasites acting as "biological weapons" have played an important role in many recent species replacements across a wide range of taxa (Prenter et al., 2004; Bell et al., 2009; Strauss et al., 2012; Dunn and Hatcher, 2015). Examples include crayfish plague (caused by the fungal pathogen *Aphanomyces astaci*) that was introduced with the invasive signal crayfish, *Pacifastacus leniusculus*, and has been responsible for mass mortality in British white-clawed crayfish, *Austropotamobius pallipes*, and noble crayfish, *Astacus astacus*, populations (Holdich and Reeve, 1991; Capinha et al., 2013). The invasive harlequin ladybird, *Harmonia axyridis*, carries microsporidia which can kill native ladybird species such as *Coccinella septempunctata* (Vilcinskas, 2015), and the expansion of the white-tailed deer in North America into territories occupied by moose and caribou was aided by macroparasitic meningeal worms carried by the white-tailed deer, *Odocoileus virginianus*, and which proved lethal to the other species (Oates et al., 2000).

Perhaps one of the best documented examples of disease-mediated native replacements is the invasion of grey squirrels, *Sciurus carolinensis*, into the UK. There were multiple introductions of grey squirrels to the UK from North America in the late 19th and early 20th century (Middleton, 1930), as well as translocations from established populations in England to other parts of the UK and Ireland. Since its introduction into the UK, the grey squirrel has "replaced" the native red squirrel (*S. vulgaris*) throughout most of England and Wales, and in parts of Scotland and Ireland (Bryce, 1997; O'Teangana et al., 2000; Gurnell et al., 2004). Scotland now has a special responsibility for red squirrels in a UK context being home to an estimated 75% of the remaining UK populations, but even here the red squirrel has disappeared from a large part of its former Scottish range in central and south-eastern Scotland. Replacement of red by grey squirrels takes the form of disease-mediated competition. Here, the grey squirrel has a competitive advantage in most habitats dominated by large seeded broadleaf trees (Kenward and Holm, 1993; Gurnell et al., 2004). In addition grey squirrels carry squirrelpox virus (SQPV) which is harmless to them but lethal to red squirrels (Sainsbury et al., 2000; Tompkins et al., 2002; LaRose et al., 2010). It has been shown that the decline of red squirrels is significantly faster in areas where grey squirrels act as carriers of SQPV (Tompkins et al., 2003; Rushton et al., 2006). Although squirrelpox was present in greys in England and Wales it was absent from populations in Scotland and it is therefore likely that the greys initially introduced in Scotland were squirrelpox-free (McInnes et al., 2009). However, the virus has been observed to be spread-

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ing northwards through England and in 2005 the first cases were reported at the England/Scotland border (McInnes et al., 2009). In response, the Saving Scotland's Red Squirrels (SSRS) project (a collaborative group consisting of government and non-government conservation agencies and land owners, see http://scottishsquirrels.org.uk/) began a programme of grey squirrel control in areas where squirrelpox was reported with the aim of preventing further spread of SQPV in Scotland. This is underpinned by field evidence that grey squirrel control can lead to a reduction in squirrelpox prevalence in residual grey populations (Schuchert et al., 2014). To prioritise grey control efforts a surveillance strategy to detect squirrelpox in Scotland began in 2005 and initially consisted of trapping and testing grey squirrels along the Scotland-England border. Testing was intensified at locations where squirrelpox was detected and in a buffer area around the furthest incidences of squirrelpox spread. From 2012–2015 a systematic testing regime was introduced in which greys were trapped and tested in alternate 10 km grid squares that cover southern and central Scotland. To get full area coverage over a two-year period the 10 km grid squares in which testing was undertaken were switched each year — but in addition continued testing was undertaken in grid squares where there had been SQPV positive results previously. Throughout the entire period deceased red squirrels found by, or sent to, squirrel officers were also tested. In this study we will develop spatial mathematical modelling approaches to assess the effectiveness of control in preventing the expansion of SQPV in Scotland.

Previous studies that have used explicitly spatial models to examine the spread of infectious disease in wildlife populations include classical Levins (1969) metapopulation approaches (Hess, 1996; McCallum and Dobson, 2002) and simulation methods with idealised spatial set-ups (Hess, 1996; Cross et al., 2005; White et al., 2014). Findings indicate that improved connectivity can enhance the spread and persistence of the disease. This suggests therefore that heterogeneity of natural landscapes and habitats may play a key role in disease spread (Ostfeld et al., 2005). A recent study by Macpherson et al. (2016), which modelled the potential spread of squirrelpox through red (only) squirrel populations on the Isle of Arran found that the disease did not spread between poorly connected regions since the high level of virulence associated with SQPV in red squirrels led to disease fade-out. This differs from the situation of SQPV in grey squirrels where infection is not associated with increased mortality and therefore can remain endemic in grey populations. This may make it more difficult to prevent the long-term spread of squirrelpox and could increase the importance of regions of poor connectivity as barriers against disease expansion. In this study, we develop spatially explicit models that include a realistic representation of the natural habitat to examine the case study of the spread of squirrelpox through the red and grey populations in Southern Scotland. Data on grey squirrel control, provided by SSRS, were included in the model to provide an assessment of how this strategy has impacted on the spread of squirrelpox in the south of Scotland and to explore if there are control strategies that can reduce its further spread. The key aims were to assess whether the SSRS control strategy can prevent the spread of squirrelpox throughout Southern Scotland and examine whether alternative control strategies could contain squirrelpox. Furthermore, the results from this case study provide a general understanding of how habitat heterogeneity and control strategies influence disease spread and persistence.

Methods

Determining potential density

The spatial model was run on a 1×1 km gridded landscape that represented the heterogeneous habitat of Southern Scotland. To define the landscape the Forestry Commission Scotland (FCS) forest inventory maps for 2011 (provided by FCS) were analysed using GRASS GIS (Version 6.4, http://grass.osgeo.org/) to determine the proportion of each 1×1 km grid square that was occupied by coniferous forest, broadleaved forest, shrub or that was inhabitable. Published densities for red and grey squirrels in each habitat type were consolidated to provide best estimate squirrel density (see Fig. 1). The gridded habitat data and best estimate squirrel density were then combined to produce potential densities for red and grey squirrels at the grid square level (Fig. 1). The grid size was chosen as it contains the core range of grey and red squirrels (assumed to be a circle of radius θ=0.15 km leading to a core range of approximately 7 ha; Smith, 1999; Bosch and Lurz, 2012) and reflects long distance movements of squirrels which can cross distances in the region of 1 km through unsuitable habitat types with no or little cover when dispersing (see homing experiments of Goheen et al., 2003).

The model

Since the number of infected individuals is low at the onset of an infectious outbreak it is essential to represent the stochastic nature of the dynamics as this can account for disease fade-out or unsuccessful invasion attempts which better reflect the population behaviour of natural systems. Therefore, we adapted the deterministic framework of Tompkins et al. (2003) to develop a stochastic model of competition and shared infection to examine the interaction between red and grey squirrels. The Tompkins et al. (2003) model system is a suitable framework as this system highlighted how squirrelpox was a key driver of the rapid replacement of red squirrels by greys and compared well to observed spatial data. The Tompkins et al. (2003) model has been successfully adapted to examine the stochastic dynamics in isolated strongholds for red squirrels (White et al., 2014; Macpherson et al., 2016). The determ-

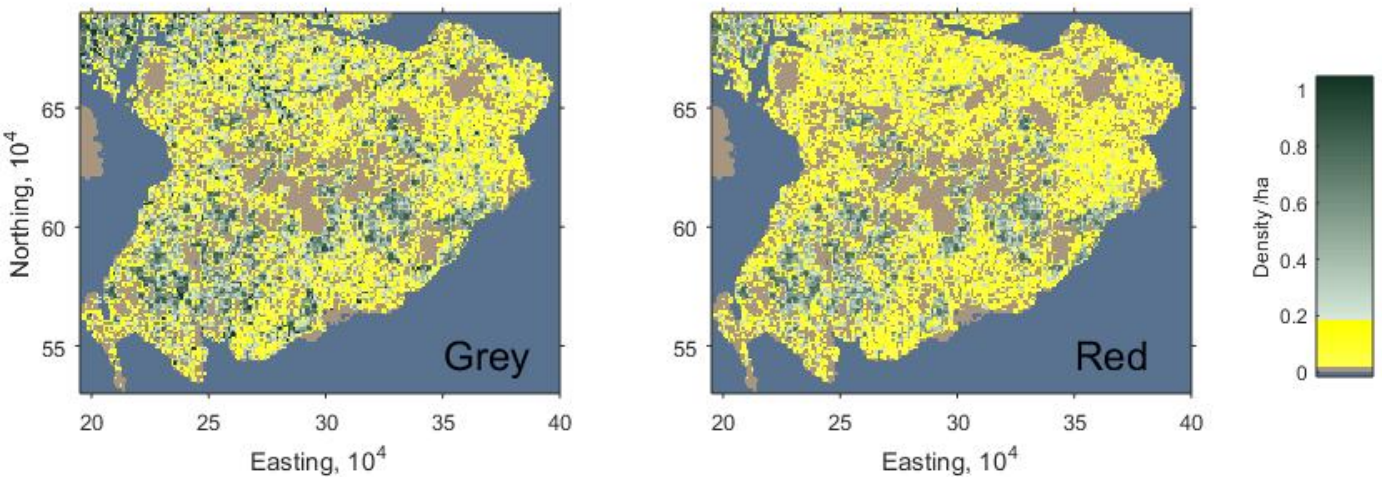

Figure 1 – The potential densities of grey and red squirrels determined from digital landcover maps and using densities estimates (ha^{-1}) in different habitats (for grey, red) as follows: broadleaf (3, 1), conifer (0.9, 0.83) and shrub (0.2, 0.2). Potential densities represent the carrying capacity of each species in the absence of the other species. Data taken from Tittensor, 1970; Gurnell, 1983; Smith, 1999; Cartmel, 2000 and Bosch and Lurz, 2012 .

inistic model of Tompkins et al. (2003) where the dynamics of susceptible and infected red (S_R and I_R) and grey (S_G and I_G) squirrels and recovered (immune) greys (R_G) are represented by the following equations:

$$\begin{aligned}
\frac{dS_G}{dt} &= (a_G - q_G(H_G + c_R H_R))H_G - bS_G - \beta S_G(I_G + I_R) \\
\frac{dI_G}{dt} &= \beta S_G(I_G + I_R) - bI_G - \gamma I_G \\
\frac{dR_G}{dt} &= \gamma I_G - bR_G \\
\frac{dS_R}{dt} &= (a_R - q_R(H_R + c_G H_G))S_R - bS_R - \beta S_R(I_G + I_R) \\
\frac{dI_R}{dt} &= \beta S_R(I_G + I_R) - bI_R - \alpha I_R
\end{aligned} \tag{1}$$

Here, $H_G = S_G + I_G + R_G$ and $H_R = S_R + I_R$ representing the total squirrel populations. Parameter values are taken from Tompkins et al. (2003) in which the two species have the same rate of adult mortality (b=0.4) but different rates of maximum reproduction (a_R=1.0, a_G=1.2) and different carrying capacities (K_R, K_G which are the potential densities in each grid cell – Fig. 1) leading to susceptibilities to crowding (q_R, q_G) (since $q = \frac{(a-b)}{K}$). The competitive effect of grey squirrels on red squirrels is denoted by c_G=1.6, whilst that of red squirrels on grey squirrels is denoted by c_R=0.61. Squirrelpox virus is transmitted at the rate β=0.55 both within and between each squirrel species. This was set using the method outlined in Tompkins et al. (2003) so that the seroprevalence of grey squirrels matched that observed from several trapping sites in Southern Scotland (see Supplemental materials). The disease-induced mortality rate for reds is denoted by α=26 and the recovery rate from infection for greys is denoted by γ=13. To generate the stochastic model the rates in the deterministic model are converted to probabilities of events that account for changes in individual patch level abundance (Renshaw, 1991). The probabilities are determined as follows:

Birth of Grey to S_G
$P(S_G \to S_G + 1) : [(a_G - q_G(H_G + c_R H_R))H_G]/R$
Natural death of S_G
$P(S_G \to S_G - 1) : [bS_G]/R$
Infection of Grey
$P(S_G \to S_G - 1, I_G \to I_G + 1) : [\beta S_G(I_G + I_R)]/R$
Recovery of Grey
$P(I_G \to I_G - 1, R_G \to R_G + 1) : [\gamma I_G]/R$
Natural Death of R_G
$P(R_G \to R_G - 1) : [bR_G]/R$
Birth of Red to S_R
$P(S_R \to S_R + 1) : [(a_R - q_R(H_R + c_G H_G))S_R]/R$
Natural death of S_R
$P(S_R \to S_R - 1) : [bS_R]/R$
Infection of Red
$P(S_R \to S_R - 1, I_R \to I_R + 1) : [\beta S_R(I_G + I_R)]/R$
Natural/diseased death of I_R
$P(I_R \to I_R - 1) : [(b + \alpha)I_G]/R$ (2)

In addition there are probabilities of individuals of each class moving to neighbouring patches. The probability of leaving the current patch for class S_G is

$$P(S_G \to S_G - 1) : [mS_G \exp(-(K_G - (H_G + c_R H_R)))]/R \tag{3}$$

here m is the long distance dispersal rate and the exponential function reflects the situation in which squirrels are less likely to disperse when the absolute density in the grid patch is below the carrying capacity and more likely to disperse when above it (and similar terms are used to represent dispersal in other classes). When an individual leaves a patch it enters a neighbouring habitable patch and therefore the density of the focal patch reduces by 1 and in the neighbouring patch is increased by 1 (the neighbouring patch is chosen at random from the 8 nearest neighbours, with appropriate weighting given to the four adjacent neighbours compared to the four corner neighbours). The long distance dispersal rate, m, when the population is at the carrying capacity, is set equal to the natural death rate, b, so that on average squirrels are predicted to disperse to a new home cell once in their lifetime.

As well as (rare) long distance dispersal, squirrel movement occurs on a regular basis within their core (home) range. This process will not lead to the permanent relocation of a squirrel to a new patch but could lead to transmission of infection by a susceptible entering an infected patch or an infected from a neighbouring patch entering the focal patch. This leads to additional probabilities of infection (for say grey squirrels) as follows

$$\begin{aligned}
&P(S_G \to S_G - 1, I_G \to I_G + 1) \\
&\quad : \left[\beta S_g \left(\theta \sum_{\text{Adjacent}} (I_G + I_R) + \theta^2 \sum_{\text{Corner}} (I_G + I_R)\right)\right]/R
\end{aligned} \tag{4}$$

The scaling for the adjacent grid patches (θ) and corner grid patches (θ^2) are determined by comparing the relative area of the patch with the overlapping areas at the grid boundary when the core range has a radius of θ km. The analogous formulation is used for red squirrels.

In equations 2–4 $R = \sum[\text{rates}]$ (the sum of the terms in square brackets) and therefore transforms the rates to probabilities. The time between events can be determined as $T_{\text{event}} = -\ln(\sigma)/R$ where σ is a random number drawn from a uniform distribution between 0 and 1 (which assumes that the time to the next event is an exponentially distributed random variable; Renshaw, 1991). The events are incremented at random with the associated probabilities updated due to changes in population density after each event. Individual simulations can be undertaken using a Gillespie algorithm (Gillespie, 1977; Renshaw, 1991) and provide information of the behaviour in a single realisation. Monte Carlo methods can be used to generate multiple simulations to assess the average behaviour and variability across realisations.

Initial Conditions

The model was initialled with observed data for the presence of red and grey squirrels in 2005 (using the National Biodiversity Network's Gateway, http://data.nbn.org.uk/). In regions where only one squirrel species was predicted the model was initialised at the respective potential density based on available habitat types (Fig. 1). In regions where both squirrel species were predicted the model was initialised by assuming that reds and greys had access to half the habitable area in each grid cell. The only caveat is that in Dumfries and Galloway the 2005 observations predicted much of the region to be absent of greys, but by 2007 to be occupied by reds and greys. For greys to spread over such an extent it was necessary to assume they were present at a low level in 2005 (5% of their potential density) in these regions. Infection was instigated to represent the first recorded squirrelpox outbreaks near Canonbie in Dumfries and Galloway in 2005. Outbreaks are also initialised near Coldstream and Eyemouth in the Scottish Borders from 2010 to reflect observed outbreaks in these regions.

Modelling Grey Squirrel Control

SSRS provided a variety of data regarding grey control strategies. This included records of trapping regimes, locations and number of animals caught; to general overviews of the percentage of large scale regions that were trapped each year. The historical and current trapping data is not in an easily usable or consistent form to generate accurate control strategies that can be used for the full region and time span considered in the model. Therefore to determine the location in which control was applied SSRS provided an estimate of the scale of trapping in 10 km

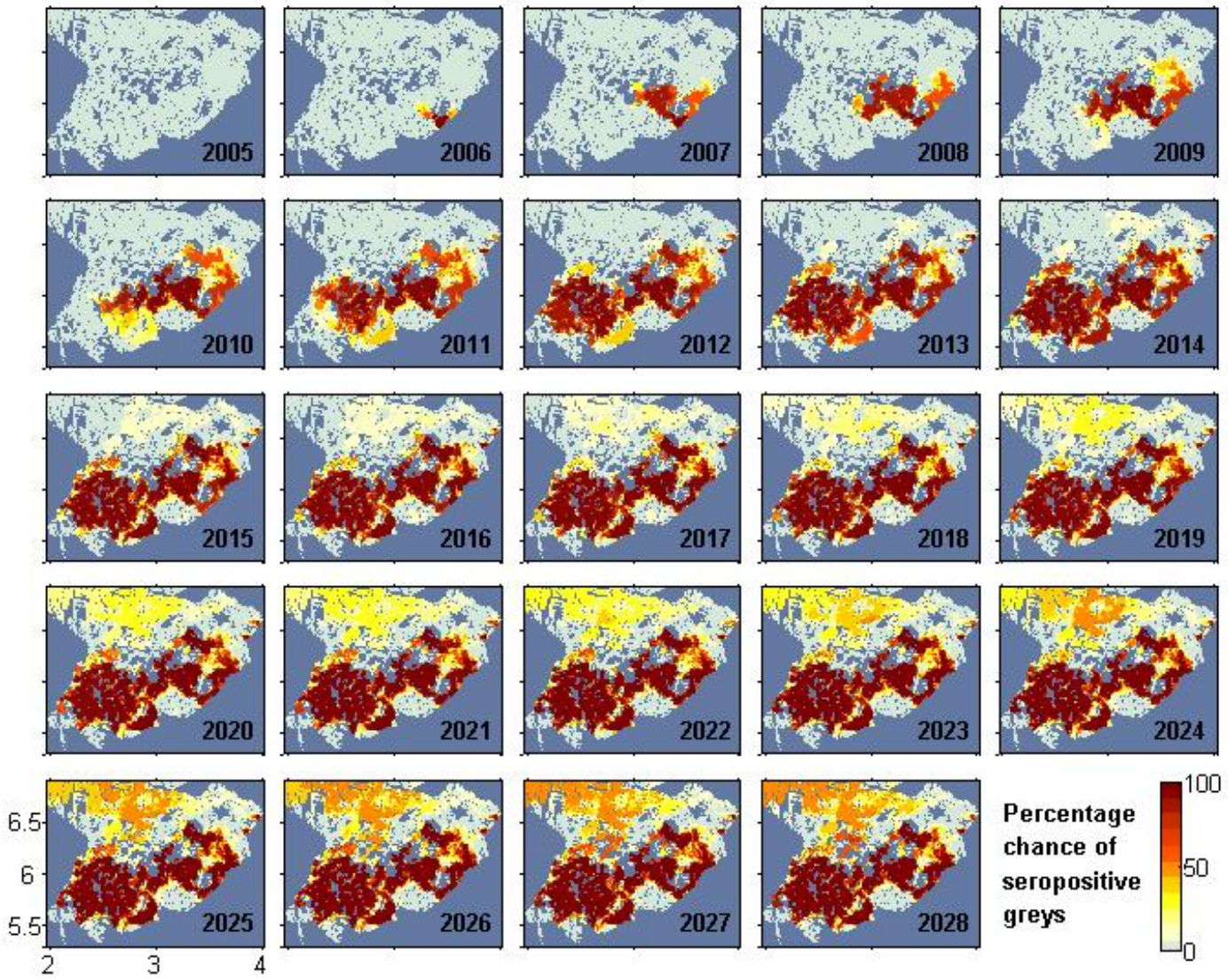

Figure 2 – The percentage occurrence of squirrelpox over time in the model. Squirrelpox is defined as occurring if seroprevalence is greater than 20% in a 1 km grid square. Easting and Northing values ($/10^5$) are shown on the bottom left panel.

grid squares for the whole of the study regions for 2005–2012 (with the 2012 level used in subsequent years). The scale ranged from 0–4 to indicate the intensity of trapping (scale 1 representing 1.25% of the 10 km grid square was trapped per annum, scale 2: 2.5%; scale 3: 5% and scale 4: 7.5%). To use this data therefore requires knowledge of the percentage of grey squirrels caught per control event. This percentage was determined using detailed trap data which was recorded by grey squirrel control officers and holders of Scottish Rural Development Grants for grey squirrel control. Here where trap location, trap days and number of greys caught was recorded the data was used to calculate the average percentage of greys caught in each 1km model gridsquare due to a control event as 40%. The grey control procedure in the model applies the reported scale of control in each 10 km grid square by locating the 1 km grid cell with the highest simulated seroprevalence (or highest density of greys if squirrelpox is absent) and removing 40% of the grey squirrels in that grid cell. The model then systematically expands the control region to neighbouring 1 km grid cells and removes 40% of greys in these grid cells until the desired percentage of the habitable region in the 10 km grid square is controlled. This procedure is followed for all 10 km grid squares in which there is control for each year.

Results

The spread of SQPV in Southern and Central Scotland without control

Model simulation were undertaken to determine the rate and extent of squirrelpox spread in the absence of any control measures that reduce grey squirrel density. These simulations provide baseline results from which the impact of control measures can be compared. They also indicate the likely progression of squirrelpox should control measures be stopped. Model simulations were replicated ten times and we present results of the distribution of SQPV in typical individual model simulations in the Supplemental Material (Fig. S3-S6 in the Supplemental Material) and the combined output showing the percentage occurrence of SQPV in Fig. 2.

The individual model simulations show that there is a rapid spread of squirrelpox from its initial point of entry in south-west Scotland and by 2012 squirrelpox is established in much of Dumfries and Galloway and the Scottish Borders (see Fig. S3-S6). The seroprevalence varies spatially from around 30%–80% and reflects the underlying distribution of squirrel density as shown in Fig. 1, where high density regions can support a higher seroprevalence in greys. In 50% of the simulations the SQPV distribution remains relatively stable between 2013 and 2028 and in particular squirrelpox does not spread and establish in the high density grey populations of Central Scotland (see Fig. S3). This pattern arises since infectious spread is limited through a band of poor (low density) habitat between Southern and Central Scotland. In the

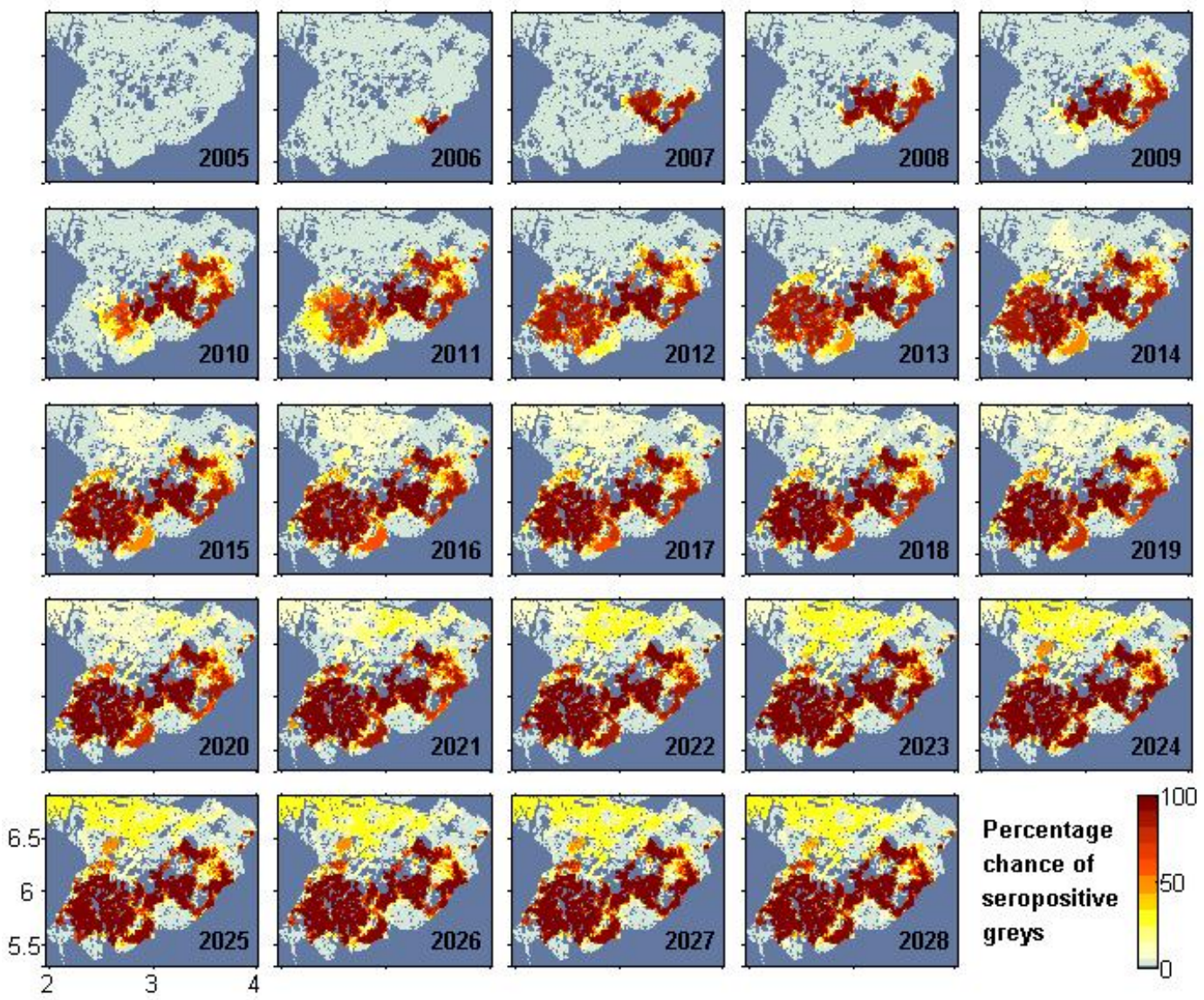

Figure 3 – The percentage occurrence of squirrelpox over time in the model with grey control (as outlined in the Supp. Info.). Squirrelpox is defined as occurring if seroprevalence is greater than 20% in a 1 km grid square. Easting and Northing values ($/10^5$) are shown on the bottom left panel.

remaining simulations SQPV successfully spreads and establishes in Central Scotland. Here the SQPV spreads through distinct dispersal "corridors" and once squirrelpox reaches Central Scotland it spreads rapidly (see Fig. S4-S6). Note that seroprevalence is either low or zero in the dispersal corridors except when (rare) epidemics occur and this highlights that relatively poor habitats can still act as dispersal corridors for infection. These results are summarised in Fig. 2 which shows the percentage occurrence of squirrelpox in the model between 2005–2028. By 2013 squirrelpox is present in all model simulations in Southern Scottish regions that can support a "reasonable" density of grey squirrels. The pattern in Southern Scotland remains largely fixed from 2014 onwards. From 2014 onwards the occurrence of squirrelpox in Central Scotland increases and by 2019 occurs in 30% of the model runs and by 2024 in 50% of the model runs. The slow spread to central Scotland is due to regions of poor connectivity in habitat between Southern and Central Scotland through which infection spread rarely occurs (see Fig. S4-S6). If the model simulations were continued for a longer time period there would be more opportunity for the (rare) spread of infection through dispersal corridors and so the probability of the occurrence of squirrelpox in Central Scotland would continue to increase.

The spread of SQPV in Southern and Central Scotland with grey control

In an attempt to prevent or slow down the spread of squirrelpox through Southern Scotland there has been a programme of control of grey squirrels coordinated by the SSRS Project. In particular, grey squirrels are trapped and removed in regions where squirrelpox outbreaks are detected. These control measures began in 2005 and have over time become more coordinated and extensive. Since 2012 they have been supplemented by systematic, strategic surveys to determine the extent of squirrelpox spread with the output from the surveys informing trapping regions. Data provided by SSRS showed the grey squirrel control officers had caught 3960 squirrels between October 2006 and August 2011 (with 1275 removed in 2010 and 1025 removed between Jan-Aug 2011). This provides a lower bound as additional grey squirrels will be caught from holders of Scottish Rural Development Grants for grey control and through trap loan schemes. The grey squirrel control procedure employed in the model (see Methods section) allowed the temporal and spatial trapping effort to be approximated between 2005 and 2012 (and the 2012 effort was applied thereafter) and this effort was included in the model. This resulted in 7550 grey squirrels being caught in the model between 2007–2011 (inclusive, see Fig. S7) which is above the lower bound set by the SSRS data. The change in the spread of SQPV in terms of the percentage occurrence of seropositive grey squirrels is shown in Fig. 3. There is evidence that the control

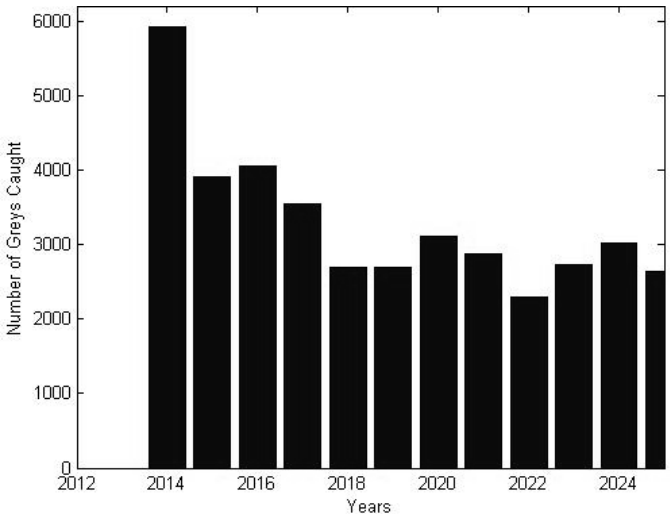

Figure 4 – The total number of grey squirrels removed each year in the model when control was targeted on dispersal corridors (see Figure S7).

measures have reduced the rate of spread of squirrelpox with the chance of SQPV reaching Central Scotland by 2028 reduced to 30% (from 50% without control). This suggests that the level of control applied by the SSRS programme reduced the spread of squirrelpox, but could not have prevented it becoming endemic across much of Southern Scotland. The model results also predict that squirrelpox can spread to populations in Central Scotland through the same dispersal routes highlighted in the model findings without control (Fig. S4-S6). These results are in reasonable agreement with observations for the occurrence of SQPV up to 2012 (see Fig. S8).

The spread of SQPV in Southern and Central Scotland with control targeted on dispersal corridors.

To assess whether the control effort could be allocated in a more effective manner to prevent SQPV spread to Central Scotland an alternative control strategy of targeting control along key corridors was tested in the model. These corridors are the regions between Southern and Central Scotland along which rare infection spread occurred (see Fig. S4-S6 and S9). In the model intensive control was applied from 2014 onwards in targeted areas (Fig. S9) with the number of greys removed per year shown in Fig. 4 indicating that initially around 6000 greys must be removed, but that this number reduces (since squirrel abundance in the control regions reduces over time) to around 2500 per year by the fourth year of control. Fig. 5 indicates the percentage occurrence of squirrelpox over time. Note that in 20% of the simulations squirrelpox spread beyond the control regions before control was applied and led to outbreaks and SQPV persistence in Central Scotland (Fig. 5). In the remaining simulations this control strategy prevented SQPV from spreading to Central Scotland. The model results suggested that a high intensity of control of greys is critical to prevent SQPV from spreading through the controlled regions since this reduced grey density to levels that restrict SQPV epidemics and therefore spread. When presented to SSRS in 2013, the results also highlighted the potential urgency of adopting such a control strategy to prevent squirrelpox spread to Central Scotland.

Discussion

This study used a spatially explicit stochastic model to examine the spread of an infection across a realistic landscape. We focussed on the case study system of the spread of squirrelpox through established red and grey squirrel populations in Southern Scotland. The findings have direct relevance to the current and future conservation strategies to protect red squirrels in the UK and also highlight general conclusions regarding the spread and persistence of infection across realistic landscapes.

The model findings suggest that there are two processes that drive the spread of infection: (1) infection is passed from individual to individual through an interlinked resident population with individuals undertaking core range movement which is not dependent on long distance dispersal of individuals, (2) dispersing infected animals move from one population to another. Process (1) primarily drives infection spread through populations in good-quality, well-connected habitats and accounts for the rapid spread of SQPV in Southern Scotland. This is directly linked to population density as a sufficient threshold density is required to support the infection. The spread of infection is (temporarily) contained by regions of poor quality habitat as here the population density is not sufficient to support the infection. However, infected individuals can spread through poor quality corridors, through process (2). This explains the delay in SQPV spread from Southern to Central Scotland, as successful spread across poor quality habitat by process (2) is rare, and also the subsequent rapid spread of infection (through process (1)) once the infection reached the good-quality, well-connected populations in Central Scotland. Note, the long-distance dispersal parameter (m) governs the rate at which "rare" infectious spread can occur across poor quality habitat. There is considerable uncertainty in this parameter and increases in the rate of long-distance dispersal will increase the distribution and speed of SQPV spread and therefore the likelihood that squirrelpox would reach Central Scotland (ongoing work is focused on determining a more accurate value for this parameter).

The model results highlight how infectious disease characteristics combine with spatial connectivity to determine the rate and extent of disease spread. In particular, SQPV is harmless to grey squirrels and this allows the infection to remain endemic. This aids the rapid spread through well-connected habitat, but also provides a continual source of infection for potential (rare) spread across poor quality habitat such as open ground with little cover. Thus SQPV is predicted to continue to expand its distribution in regions occupied by grey squirrels. This contrasts with previous modelling results that have examined the spread of SQPV in red squirrel populations (White et al., 2014; Macpherson et al., 2016). The key difference is that SQPV causes high levels of disease induced mortality in red squirrels. In red squirrel populations the disease is predicted to cause a local epidemic followed by disease fade-out (White et al., 2014; Macpherson et al., 2016). Therefore, the disease is not predicted to spread extensively through red squirrel populations and this confirms predictions by Duff et al. (2010) who argued that SQPV would burn itself out in red-only squirrel populations due to the high rate of disease induced mortality. Our results on realistic landscapes also support model studies that use idealised spatial set-ups (Hess, 1996; Cross et al., 2005; Macpherson et al., 2016). Here chronic, low virulence infection can spread extensively as it can be supported in local populations and thereby provides the opportunity for the rare spread of infection between poorly connected populations. In contrast, acute, highly virulent infections only spread locally as the disease fades-out before spreading to neighbouring populations.

The model results in this study also have direct implication for conservation management. In response to the first outbreaks of SQPV in Scotland the Saving Scotland's Red Squirrels project implemented a strategy of grey control in regions where SQPV was detected (SNH, 2006). This strategy did not succeed in preventing the spread of SQPV throughout Southern Scotland (see Fig. S8). The model results indicate that control of grey squirrels in response to SQPV outbreaks may slow the spread of infection, but would not prevent it as in the well-connected habitat in Southern Scotland there are many alternative routes for squirrelpox spread that circumnavigate control areas. Also, detection of the squirrelpox may lag behind the spread of the disease and so SQPV spread is likely to be more extensive than that observed from field testing. This highlights the difficulty in trying to prevent disease spread in a reservoir population.

The model findings also indicated that the spread of infection may be limited along key corridors due to regions of poor habitat (and hence low squirrel density) that offer poor connectivity between regions of high abundance where the infection can persist in the long-term. The model results suggested that applying grey control at a sufficiently high

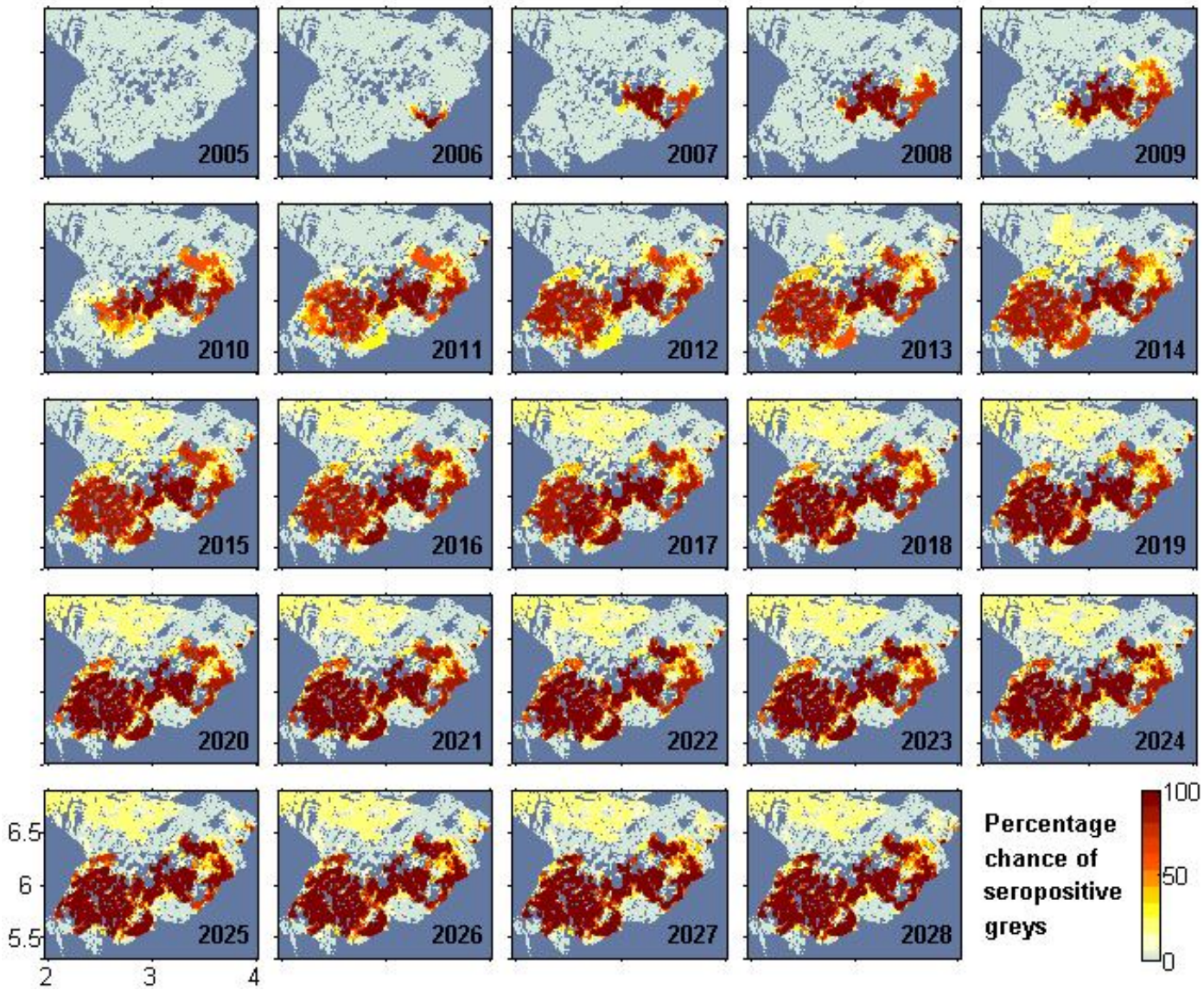

Figure 5 – The percentage occurrence of squirrelpox over time in the model with grey control targeted on the dispersal corridors outlined in Figure S7. Squirrelpox is defined as occurring if seroprevalence is greater than 20% in a 1 km grid square. Note, outbreaks in central Scotland occur in 20% of model simulations as the disease spreads beyond the control regions prior to control being applied (see year 2013). Easting and Northing values (/105) are shown on the bottom left panel.

intensity in targeted regions could prevent SQPV spread from Southern to Central Scotland (Fig. 5). However, a compounding factor is that the model only considers habitat as defined by the forestry inventory data set and so does not consider small (less than 0.5 ha) woodlands or urban habitat. This additional habitat may enhance dispersal, increase the spread of squirrelpox and lead to difficulties in isolating dispersal routes in the field. In September 2013 the SSRS steering group considered targeting control on key dispersal corridors. A wide ranging discussion involved a consideration of the likelihood of success and the resource implications of high intensity control along the broad front between Southern and Central Scotland (where the model indicated that the pinchpoints were located). It was noted that this new strategy would divert resources from protecting populations in areas of Southern Scotland where there are still population of red squirrels that are under threat from greys in which squirrelpox is endemic. To confound matters further squirrelpox was detected in isolated grey squirrels sampled in Central Scotland at the end of 2013 and in 2014 (Fig. S10), suggesting that the window of opportunity may already have passed. Therefore a targeted strategy of grey control at pinchpoints between Southern and Central Scotland was not implemented (but ongoing work is examining the utility of this approach to prevent SQPV spread to Northen Scotland). The SSRS red squirrel conservation strategy for Southern Scotland was therefore modified (SNH, 2015) with the focus changing from preventing squirrelpox spread to protecting red squirrels in Priority Areas for Red Conservation (PARCs). Evidence from field studies (Chantrey et al., 2014; Schuchert et al., 2014), conservation groups (Gurnell et al., 2014) and modelling (White et al., 2014, 2015) that has examined the viability of red squirrel strongholds (priority areas) indicate that reds can survive if control prevents grey squirrels from establishing in the priority areas even when squirrelpox is endemic within grey populations (Lurz et al., 2015). However, studies do suggest that the adjacent endemic grey squirrel populations can infect the protected red population. This can lead to periodic outbreaks of squirrelpox in the red squirrel populations which causes a population crash, followed by disease fade-out (as the low density following a crash cannot support the disease) and a return to pre-infection population level in red abundance (Chantrey et al., 2014; White et al., 2014; Lurz et al., 2015). In Southern Scotland the PARCs reflect areas where there has been success in retaining viable red squirrel populations (through the continual control of grey squirrels) despite the presence of grey squirrels and squirrelpox outbreaks and where there has been significant, longstanding local effort for red conservation (SNH, 2015). Spatial modelling suggests that in principle the PARCs focus can be successful in protecting red squirrels (White et al., 2014, 2015) and ongoing work is adapting the models to determine the optimal effort of grey control required within and around them. Beyond Southern Scotland

there is still a focus on restricting the spread of squirrelpox to prevent it from reaching the established, red squirrel only, populations in Highland Scotland. Here the model system can be used to isolate the key corridors along which squirrelpox spread is limited between Central and Highland Scotland and therefore where targeted control could still be effective.

This study has shown how mathematical modelling techniques that include a realistic representation of the heterogeneous spatial structure of natural systems can inform and help direct conservation strategies to protect endangered species. The techniques are easily adapted to represent different systems and since the stochastic population and disease dynamics are underpinned by classical deterministic modelling frameworks the results are readily interpreted and applicable in general. Our results therefore show how stochastic modelling frameworks are essential tools for conservation management.

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Supplemental information

Additional Supplemental Information may be found in the online version of this article:

Equation S1 Calculation of the disease transmission coefficient.

Table S2 Trapping data for sites in Southern Scotland, carrying capatities, seroprevalence and transmission coefficients.

Figure S3-S6 The spread of squirrelpox from its initial introduction in 2005 in a single model simulation.

Figure S7 The total number of grey squirrels removed each year in the model (for a typical model realisation).

Figure S8 The recorded incidence of squirrelpox in the south of Scotland in 2010 and 2012 (provided by SSRS).

Figure S9 Map of regions in which the targeted control procedures was applied to prevent SQPV spread along key corridors connecting Southern and Central Scotland.

Figure S10 The recorded incidence of squirrelpox in the south of Scotland from 2012 to 2014 (provided by SSRS).